Pergamon

Biomass and Bioenergy Vol. 11, No. 1, pp. 11–32, 1996

PII: S0961-9534(96)00006-2
0961-9534/96 $15.00 + 0.00

THE CROP PHYSIOLOGY OF *HELIANTHUS TUBEROSUS* L.: A MODEL ORIENTATED VIEW

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(Received 19 January 1996; accepted 22 January 1996)

Abstract—Information from the literature is discussed to define guidelines for a model of the growth and development of *Helianthus tuberosus* in non-limiting conditions. Dynamics of the leaf area index and the distribution of growth between structural growth and reserves (in stem, stolons and tubers) are the most crucial processes. Phenological events like stolonisation and tuberisation seem to be controlled mostly by plant growth. In this hypothesis, the main true developmental event is flower initiation. This last event is the main physiological shift determining the tuber yield and quality through change in assimilate distribution caused by saturation of aerial sinks. Directions are proposed for future research efforts aimed at improving the insight into crop physiology and gathering missing data for model parametrisation. Response of growth to water shortage, and nitrogen nutrition will be the next crucial questions to model the crop under field conditions. Copyright © 1996 Published by Elsevier Science Ltd.

Keywords—*Helianthus tuberosus* L.; Jerusalem artichoke; development; fructan; growth; model; reserves; tuber.

NOMENCLATURE

ABA	Abscisic acid
cv	Cultivated variety
DP	Degree of polymerisation
DW	Dry weight
F	Fructose or fructosyl
FEH	Fructan-exo-hydrolase
FFT	Fructan-fructan-fructosyl transferase
FW	Fresh weight
G	Glucose or glucosyl
HI	Harvest index
IAA	Auxin (β-indolylacetic acid)
LAI	Leaf area index (m^2/m^2)
MCPA	2-Methyl 4-chlorophenoxyacetic acid
NAA	α-naphtylacetic acid
PAR	Photosynthetically active radiation (wavelength: 400–700 nm)
PFD	Photosynthetic photon flux density (corresponding to PAR)
SS	Sucrose synthase
SST	Sucrose-sucrose-fructosyl transferase
TIBA	2,3,5-Triiodobenzoic acid

1. INTRODUCTION

Helianthus tuberosus (Jerusalem artichoke in English, topinambur in German, topinambour in French, aardpeer in Dutch, pataca in Spanish) almost disappeared as an agricultural crop. Recently, its reintroduction has been studied in many countries for a possible re-development,[1] particularly in relation to its hardiness[2] and low production costs.[3,4]

At present, *H. tuberosus* is mainly considered as a biomass crop for ethanol production[5,4] because it commonly yields around 7 and potentially up to 14 tonnes of carbohydrate per hectare (t ha^{-1}).[6,7] Other industrial uses for the crop are: paper pulp or fuelwood from stems,[2,8] methane production with various plant parts,[9] acetone–butanol–ethanol production from tubers or whole plant,[4] hydroxymethylfurfural as a basic molecule for the chemical industry.[10] A large part of scientific literature deals with industrial transformations[5,10] or optimisation of combined production (like alcohol + yeast).[4,5] Besides these industrial uses, *H. tuberosus* is still of interest as a food for human consumption (including beverage alcohol and beer),[2,11] feed from tubers,[7,8] or forage from aerial parts.[8,12] High quality protein concentrate can be extracted from the leaves.[13] All plant parts can be ensiled before animal or industrial use.[12,14] Fructans (linear polymers of fructose) represent the largest part of the biomass.[15] *H. tuberosus* is therefore a source of fructose syrups for the food industry,[1] as well as fructans for medical or dietetic[16] purposes. It is also used as anti-erosion

protection to fix terraces or unstable sand,[17] or as a barrier against fire in large forests.[8]

The multiple use of the crop justifies a survey of its potential productivity as well as its crop quality (amount of sugars, degree of polymerisation of fructans, chemical composition). A few bibliographies[18] or reviews on *H. tuberosus* have been published already, either giving a general introduction to the crop and its various uses,[16] or focusing on *H. tuberosus* as a weed in America,[19] or oriented towards industrial uses and transformations.[20] A few recent proceedings deal specifically with *H. tuberosus*,[21,22] or fructans and fructan-containing crops and their uses.[11,23] However, no recent synthesis about the physiology of this crop has been found in bibliographical data banks.

Simulation with dynamic models may be helpful for quantitative estimations of yield and quality of a new crop when field references are lacking.[24] It is also helpful to identify the most strategic orientations for new research. A few attempts already deal with *H. tuberosus*,[25-28] but these models still need to be improved for broad scale relevance.[29] Static models have been proposed,[30] but they are only of descriptive interest.

This review article has two objects: (1) to provide an updated synthesis of the physiology of *H. tuberosus*, (2) to identify the most important crop physiology traits for modelling purposes. This conceptual framework could help to define directions for research and for improving agricultural practices. It gives guidelines for modelling the growth and development of this crop, with simulation of quantities and qualities of the different plant parts. This article is a selective review, because of the amount of literature (more than 1100 references) based on a detailed bibliographical analysis.[31] References were obtained from public data bases: AGRICOLA (U.S.D.A., U.S.A.), CAB (Commonwealth Agricultural Bureau, U.K.) CURRENT-CONTENTS (I.S.I, U.S.A.), PASCAL (C.N.R.S., France), from the private data base of Dr Fuchs (Agricultural University, Department of Phytopathology, Wageningen, The Netherlands) and from bibliographies cited in publications.

2. GENERAL CROP FUNCTIONING

H. tuberosus is an annual crop first described more than two centuries ago, when it was introduced into Europe from North America.[8] It probably results from crossing between di- and tetraploid *helianthi* and can be crossed with other species of this family.[32] The genetic variability is large, particularly among the late varieties.[33] It normally sprouts in spring, develops a strong aerial structure, often multistem and ramified, and then tuberizes as aerial parts die off.[8,34] It can also be cultivated as a multiannual or perennial crop, from its volunteer regrowth.[2,12] The following development stages are commonly identified: emergence, stolonisation, tuber initiation, flower initiation, tuber filling, flowering, fruiting and maturity. Depending on the variety (cv), the growth cycle can be as short as 100 days[7] or as long as 9 months.[35] Short-cycle varieties are described as 'early' and they flourish in summer, whereas 'late' varieties often produce buds but no flowers. A typical trait of *H. tuberosus* is the temporary storage of a large amount of assimilates in the stem,[36,37] and another is the glucidic metabolism based on fructans (fructose polymers).[15,38]

Yields of crops have been measured in various conditions and show a large variability. Total dry weight (DW) (total biomass) of the crop ranges from 6–9 tonnes per hectare (t ha^{-1}) in poor conditions to 20–30 t ha^{-1} in good conditions.[6,39] Similarly, production of tubers ranges from 30 t ha^{-1} fresh weight (FW) or 4–6 t ha^{-1} DW,[39,40] to 50–70 t ha^{-1} FW[8,30] or 10–15 t ha^{-1} DW.[41,1] Most of the published data do not make it possible to analyse the reasons for these variations, but it must be noted that the lowest tuber yields are often caused by an excessive lateness of the cv relative to the local climate.[3,40]

Total productivity, and often tuber yield, are correlated strongly (almost linearly) with the amount of radiation intercepted by the crop.[28,42] This in turn depends on the duration of the crop,[33] the dynamics of leaf area index (LAI),[33,43] the photosynthetic performance of the canopy and the cost of synthesis of various biological compounds.[24,28] Tuber yield depends finally on the distribution and the re-distribution of assimilates between the various compartments of the plant. This last point is illustrated by the strong variation in the harvest index (HI) of the crop, generally higher in early (0.6 to 0.78) than in late cv (0.5 to 0.55) in spite of the similar total productivity per unit of time.[37,43]

Consequently information is organised further in this article according to the following

progression: total growth (duration of the crop, dynamics of leaf area, photosynthesis and radiation use efficiency, chemical composition), development of the crop (stolonisation and tuberisation, flowering), characteristics induced by fructan metabolism (including quality of sugars), distribution of growth and reallocation of compounds (growth of aerial structures and roots, growth of stolons and tubers, competition between organs).

Most of the existing data has been gathered from experiments in conditions close to optimum, that is where the crop did not suffer from shortage of water or nutrients. Thus most of the following concepts apply first to these 'non-limiting' conditions (even if they remain partly valid when some limitation occurs). Some information about the complementary impact of shortages and stresses is presented in a separate paragraph.

3. TOTAL CROP GROWTH

3.1. *Establishment and duration of the crop*

Tubers are dormant for some time after maturity, and *H. tuberosus* is thus a model-plant for many studies on dormancy.[44,45] Dormancy (a strongly cv-related character) is normally broken by cold temperature (5°C or less),[46,47] and has no more effect at the time of planting or regrowth. If dormancy is not suppressed, and only in this case, the fact that the tuber is placed in conditions adequate for germination results in the growth of a new tuber (or 'secondary tuber') at the end of the dormant tuber. This is a kind of 'secondary bulking'. It can repeat itself many times until reserves are exhausted. When dormancy is not suppressed completely, it results in germination without elongation and temporary rosette-like plants.[3] This behaviour can be avoided with long daylength treatment[48] or, as well as dormancy, by applying gibberelin.[46]

Planting takes place in autumn or spring,[35] generally in ridges. Success in emergence is generally high (98%–100%),[12] but is sometimes lower in warm climates, probably because of temporary dry conditions.[49] A threshold temperature between 5° and 2°C is found for sprouting and partial anoxia also limits respiration and sprouting.[16,50] Emergence begins generally 3–5 weeks after planting, with a difference of a few days between cv,[8,51] but sometimes as early as 2 weeks after planting.[30] It is generally concluded that emergence depends mainly on temperature regime,[52] as confirmed by the positive effect of covering the soil with polythene film.[3] Tuber pieces sprout as well as entire tubers.[8,45] Reduced emergence sometimes reported in such cases is certainly the result of excessively small pieces.[2,3] Optimum tuber piece size (i.e. the smallest size with maximum emergence) is around 40–60 g FW[16] with diameter larger than 35 mm.[3] An increase of the seed-tuber size often increases the final yield, but can decrease the overall biomass balance if the yield increase is lower than the supplementary seed-rate.[50,53] The most obvious effect of larger seeds can be seen in the increase of stem number and consequent early LAI growth.[8,53]

Pre-sprouting can have some positive effect on emergence, early growth and yield, if no damage to sprout occurs during planting.[3,54] Early growth often proceeds slowly in spite of the availability of sugars in the tuber seed, and is probably limited by low temperature with variations between cv.[39]

Besides nutrient and water availability (the effect of soil is mainly related to water regime[16]), soil conditions (pH, texture) have slight or no impact on crop growth.[8,53] Only wet or hydromorphic soils must be avoided because of reduced emergence, enhanced diseases, poor tuberisation, and rotting of tubers.[2,49] Early cvs are often more sensitive to exceedingly dry or wet soils.[49]

In non-limiting conditions, the plant population for maximum tuber yield is around 5–8 per m^2, somewhat higher than the conventional values (3–4 per m^2).[14,55] The input of biomass corresponding to seed-tuber is increased with a higher seed rate. Thus the planting density to obtain an optimum biomass balance of the crop is somewhere between standard density and density for maximum tuber yield. Increase in harvest index explains a large part of the improved yield with a higher planting rate.[56] A still higher density (11 per m^2) increases the production of aerial parts.[12] As water is often the most limiting factor, planting density has to be adapted to water resources and regime.

Senescence is defined as death of the whole aerial stem, a few days or weeks after the whole foliage is dry.[52] It occurs normally some weeks after flowering or flower bud appearance. Frost sometimes kills late cv. Very high planting rates (or weak growth),[57] short daylength,[58] and stresses accelerate senescence. In contrast, cold

temperature slows down senescence.[52] The crop is considered as mature when the aerial parts are totally dry. Then tuber DW is at its maximum.[8,30]

3.2. *Dynamics of leaf area index (LAI) and light interception*

In normal crop conditions, the sum of the area index of green and dead leaves can be high (up to 10 or more).[12,27] Stem number, which partly depends on seed-tuber size, plays some role in the early growth rate of LAI.[43,59]

At the canopy level, the rate of LAI increase correlates better with temperature than with time[25,26] and depends on cv.[43] In spring, the growth of leaves is more limited by temperature than by assimilates or reserves.[34] Early LAI growth[26] or early leaf number[59] fit best with a multiplicative function of accumulated degree-days. After canopy closure, LAI increases almost linearly with degree-days.[25] Foliage growth response to temperature is indeed not fully linear and needs a more detailed definition of effective temperature,[29] even for phyllochron description.[59] Flowering (or flower initiation) marks the end of LAI increase and foliage senescence is correlated with flowering date[39] or temperature sum since emergence for early cv.[59] An analytical model of LAI increase has been described; it is driven by temperature and simulates in detail the morphology of the plant.[59] But it needs thorough re-parametrisation between experiments because of various interactions between all components of the LAI (number and size of organs).[27,59] Another modelling approach of LAI increase is based on empirical allocation of dry weight increase to leaves combined with a fixed value for specific leaf area.[28] Yet this last parameter proves to be variable, increasing with time,[26] and varying between locations.

Senescence of the lower leaves during crop growth is caused by strong shading[51] and high temperature (increased respiration).[34] Accordingly, high planting density promotes canopy senescence.[57] Leaves separated from the plant survive longer than leaves attached to the plant[60] and late nitrogen supply keeps the foliage green for a longer time (but without gain in tuber yield).[3] Final senescence of the foliage is consequently ascribed to competition between plant parts particularly for nitrogen. Leaves are not very sensitive to frost during early growth, but more in autumn, and foliage of late cv is often killed by frost.[39,52]

At canopy level, Beer's law is generally used to represent light interception or absorption as a function of green LAI.[42,57] Light extinction coefficient (k) varies from 0.78 to 1.01 when fitted to experimental data, depending on cv.[25,37] Indeed, a sigmoid curve would better fit to a low LAI, probably because of some row effect.[25] The planophile[26] leaf angle distribution is almost uniform between 0 and 55° and leaf azimuth distribution is nearly perfectly random.[61,62] Optical properties of leaves are similar to those of common crops.[26,61] Albedo of the canopy (around 5% in PAR wavelength) is slightly increased (plus 1%–2%) during flowering.[61] When introduced in a light interception model these data yield a k-value between 0.95 and 0.76 when solar elevation is between 30 and 90°, close to experimental results.[62] Evolution of the light interception coefficient is also directly modelled as a logistic function of the temperature sum during increase and a linear function of the temperature sum during decrease.[28]

3.3. *Photosynthesis and total growth*

The chemical pathway and light response curves of photosynthesis at leaf level are typical of C3 plants.[63,64] There are large differences between cv for the maximum CO_2 assimilation rate for a single leaf (29–40 micromol CO_2 m^{-2} s^{-1}), but without correlation with tuber yield.[64] The value of the maximum assimilation rate is correlated strongly with the N and chlorophyll content of the leaf.[64] Photochemical efficiency, similar to sunflower,[64] can be close to that of some C4 plants.[63] It still decreases after initiation of flowers,[63] or at the time of strong tuber filling when leaf nitrogen is translocated to the growing tubers.[64] Leaf transpiration parallels photosynthesis when PFD is between 400 and 1200 micromol m^{-2} s^{-1} (giving a stable water use efficiency) and is stable for lower irradiance.[64] The compensation point for photosynthesis[64] is at 55–60 micromol m^{-2} s^{-1}.

At canopy level, the conversion ratio of intercepted (or absorbed) radiation to dry matter (or radiation use efficiency, RUE) has been estimated.[26,28,42] A better conversion at tuber filling time is sometimes recorded.[26,37] This is ascribed to the lower energetic cost of chemicals synthesized, fewer losses when temporary storage is bypassed, or stimulation of photosynthesis when the storage processes in tubers become more effective.[65,66] Incoming light is intercepted strongly in the upper layers, because

of the leaf angle, with possible saturation of the leaf photosynthetis at high PFD.[62]

Information about respiration is very scarce, thus a complete carbon balance of a crop is rare.[57]

3.4. *Chemical composition*

Owing to the large transfers of carbohydrates, proteins and minerals between plant parts during growth and particularly at maturity,[17,67] any analysis would have to refer precisely to the development and growth stage. Moreover, variations are induced by differences in cv (for instance, for tuber mineral content[68,69]) or in analytical procedures, particularly for carbohydrates. Information is often incomplete and a comparison between references is hence somewhat hazardous and therefore not given in detail.

When compared to stems, leaves (20%–30% of aerial DW) are far richer in minerals (14%–19% ash against 2.5%) and proteins (13%–24% against 1.4%–6%), poorer in lignin (3.8% against 6.2%) and carbohydrate, and equivalent in fibers.[13,70] The fructan content of stems is the most variable characteristic because of the temporary reserve storage. Leaves are able to accumulate excessive amounts of Ca, and to a lesser extent Cl, K and Mg,[50,71] with damaging consequences for their quality as forage.

In tubers, the dry weight varies between 12% and 24% of fresh weight,[39,49] with a special case for dwarf cv (25%–26%).[40] The water content of tubers (and thus apparent DW percentage) varies with soil moisture.[17] The DW percentage[49,72] is also increased by higher soil mechanical resistance, higher latitude, and stronger insulation late in the season, as plant density or N fertilisation have no or negligible effect.[50] Carbohydrate (hexoses and fructans: 60%–80%), then nitrogenous compounds (6.5%–16%, often over 10%) and ashes (5%–7%) represent most of the dry weight. Tubers of *H. tuberosus* are rich in minerals, particularly potassium.[50,68] The remainder is mostly cellulose and hemicellulose (10%–13%), lipids (0.15%–0.7%), lignin (0.6%), and organic acids.[1,73] Thus sugars represent 10%–20% of the fresh weight.[8,15] As fructans represent by far the largest part of DW, sugar and dry matter percentages are correlated strongly at harvest.[39,72] After the beginning of tuberisation, the percentages of dry weight and sugar both increase during growth.[27,54] Accordingly, the percentage of minerals decreases, while evolution is more variable for nitrogen.[69,74] At a similar physiological age, DW and sugar percentage vary between cv: lateness, darker colour, and tapered form are generally positively correlated with higher DW, DP and sugar content.[39,52] The percentage N in tubers shows slight or no significant variation between cv, and N/DW ratio is rather stable[50,74] even if big tubers are richer than small tubers.[74]

4. DEVELOPMENT

Development stages for *H. tuberosus* are rarely completely and clearly defined.[75] Thus comparison between references must be made with caution. Moreover, dates for subterranean development are not very precise because observation of underground parts cannot be easily carried out frequently.

4.1. *Subterranean development: stolonisation and tuberisation*

Stolonisation, defined as appearance of the first stolons,[75] is early during the plant cycle but references are inconsistent, varying from 9–10 days to 2 months after emergence.[52,67,75,76] Appearance and ramification of stolons proceeds for a long time, even after tuberisation has begun, but with a reduced growth rate and more ramification of existing stolons than creation of new stolons.[75,76] Stolonisation is independent of daylength[58] and planting date.[45] An analytical description of stolon number has been proposed, but this number can vary through various processes.[26,75] An increased planting rate results in a delayed appearance of and inhibited ramification,[77] and weather conditions affect elongation[45,75] of stolons. Stolonisation is probably driven by the total growth of the plant, and never seems to limit the final number of tubers.[26,65]

Tuberisation, one of the most studied traits of *H. tuberosus* physiology but still a much debated question, is generally defined as the appearance of the first tubers (i.e. swollen stolon). This concept still varies between authors and often the initiation of first tubers (sometimes called 'tuberisation' or 'tuber set'), and the subsequent—less or more delayed—tuber growth (sometimes called 'bulking') are distinguished.[75] Most references deal with the first phenomenon. In the literature, phenomena are sometimes considered at individual organ

level and sometimes at plant level (groups or cohorts of organs) thus comparison between references is not straightforward. Most research has been carried out with late cv showing a first phase of appearance and slow growth of some tubers; this is followed by a second phase of stronger growth of the first tubers ('bulking' at organ level) and the simultaneous appearance and fast growth of new tubers. At whole plant level, the first phase is called 'tuberisation' or 'initiation'. The second phase called 'bulking' or 'filling' covers initiation of new tubers. The first tubers appear 5 to 13 weeks after emergence, depending on cv and environmental conditions, some days or weeks after stolonisation.[12, 67] Tuberisation can begin as early as the beginning of June (even for late cv).[25, 78] It is sometimes correlated with the temperature sum after crop emergence, whatever the earliness for flowering.[75, 78] Contrary to what is seen with potato, there is no clear separation between tuber induction and tuber growth; both occur simultaneously and new tubers can appear at any moment until late in the season.[45, 75] In the beginning, tuber initiation is reversible: swollen stolons can recover and regrow anew as stolons,[76] as confirmed *in vitro*.[44] With prolonged inducing conditions this becomes irreversible.[79] The first tubers are often elongated and found at the end of old and long stolons; last formed tubers are rounder and set on short stolons.[52] Under very high planting rates, no or few stolons appear and the roots or stem can swell instead of normal tuberisation.[77]

The initiation of tubers is often correlated with short daylength, whatever the temperature regime, but there are large differences in varietal responses.[44, 58] These results come from controlled daylength experiments,[48, 80] sometimes coupled with grafting experiments. From such data, it is concluded that some 'stimulus' (non specific to cv and not even to *H. tuberosus*) is transferred from upper to lower plant organs.[36, 81] Grafting of early cv on late stock results in strong tuberisation (contrary to inverse grafting)[80] thus early cv would always be 'induced'. A photoperiodic signal is detected by mature upper leaves and the response is proportional to the number of treated leaves.[80]

Other experiments show that cool night temperatures are more effective than short photoperiods in inducing tuberisation, and this 'induction' is also transmissible by grafting, with only a secondary and additive effect of daylength.[44, 58] Plants from aged tubers show an earlier tuberisation.[44]

Assimilate availability and total growth of the plant also play some role in tuberisation. Reduction of photoperiod under a certain threshold or shading of whole plant leaves decreases tuber formation in comparison with shading limited to upper leaves.[48, 82] On the contrary, high light intensity promotes tuberisation even in 'non inducing' conditions.[44] Moreover in daylength experiments (most of them being carried out indoors, with low PFD levels), all plants tuberize in the end whatever the treatment.[44, 58] In field experiments tuberisation often occurs under long (even increasing) daylength. This applies not only to early cv (supposed daylength neutral) but even to photoperiodic sensitive cv, also with some positive role of early planting date and large plant size.[78] Nevertheless, tubers formed under long days are smaller, with lower growth rate, and are less typical than under short days.

Action of natural growth regulators (plant hormones) is likely in tuberisation because (a) photoperiod is sensed far from the stolons, (b) 'induction' can be transmitted through a graft, (c) hormones are active in morphogenesis and there are morphological changes at tuberisation,[52] and (d) any bud of any organ (including aerial ramification) of an induced plant can produce a tuber when kept in darkness and damp.[83, 81] Dormant tubers always show a secondary bulking instead of sprouting and primary bulking tubers are always dormant. Thus primary and secondary bulking can be seen to be similar as far as bud physiology is concerned. Action of growth regulators was investigated *in vitro* to find an hormonal determinism of tuberisation with trials to regulate bulking or sprouting on dormant or non dormant buds.[44]

Various attempts have been made to identify a 'tuberisation factor'. An unknown compound extracted from tubers promotes tuberisation of explanted buds after inhibition of their growth in length.[44] Various chemicals with growth regulating action have been applied to plants: auxin or homologues,[80] maleic hydrazide,[46] kinetin,[84] etc. Variation of natural abundance of growth regulators was also investigated.[58] The most striking results have been obtained relative to gibberelins and abscisic acid (ABA). Gibberelins inhibit tuberisation, prevent existing tubers from bulking and induce the growth of the stem.[36, 44] They stop dormancy (but are

less effective than cold treatment), also appear at sprouting and induce strong invertase activity, preventing sprouts from accumulating sucrose. On the contrary, ABA or antiauxin compounds (TIBA) or anti-gibberellic growth regulators stop stem elongation, accelerate tuberisation and increase the sugar yield.[36, 80] Short day treatment combined with any chemical spraying stimulates tuber initiation but daylength remains more efficient than any chemical alone.[84] High sucrose concentration plus ABA[83] or an antigibberellic compound[79] induce tuberisation *in vitro*, as does a still higher sucrose concentration alone, or any strong increase of osmotic potential of the growth medium. Thus the high ABA content found in very young tubers would simply be a consequence of the osmotic shock inducing tuberisation.[85]

To summarize, various hormonal schemes consistent with photoperiodic sensitive behaviour have been proposed[46, 58, 79–81, 86] for *H. tuberosus*, based on the change of balance between gibberellins and ABA in leaves, and auxin and cytokinins in roots and stolons. Tuberisation could be caused by the concentration of an hormonal compound (similar to cytokinins) in apices of stolons when their growth is inhibited.[44, 58] But the importance of assimilate availability is also often underlined.[44, 58]

4.2. *Aerial development: flower initiation, flowering, fruiting*

Induction of flower initiation and flowering have mostly been studied to carry out crossing of various cv.[87, 88] The time-span between emergence and flowering also defines earliness, one of the most recognised parameters for classification of cv.

Morphologically, floral initiation results in the swelling of the terminal bud with more foliar primordia,[78, 82] soon followed by the differentiation of floral pieces within the bud. Floral initiation depends mostly on temperature sum after emergence in the earliest cv and photoperiodism (short daylength) in late cv, as shown in daylength or planting date experiments.[89, 90] Indeed, only the photoperiodic mechanism (i.e. late cv) has been extensively studied. Flowering of photoperiodic cv before summer solstice in low latitudes shows that flower initiation is not sensitive to the direction of daylength change.[91] The effect of daylength is quantitative as the efficiency of short day treatment (number of initiated flowers per plant) depends on its duration and increases with the number of days of treatment.[73, 89] Moreover, the duration of treatment (number of days) must be proportional to daylength (i.e. longer with longer days[88]), and the effect of treatment depends on the number of treated leaves.[80, 87] The exact daylength threshold is rarely quantified.[78] The day–night temperature regime also plays some role.[91] During a 'juvenile phase' concomitant with opposite phyllotaxy, the young plant is not sensitive to flower initiating stimuli.[57, 82] As a consequence short day varieties generally do not initiate flowers in spring.[48] Flowers are initiated at the end of ramification 1 to 2 weeks after initiation of flower on main stems.[78] Upper developed (and middle) leaves are the sensitive organs for photoperiodism.[80, 87] Removing upper leaves always speeds up flowering (even with early cv);[89] thus it would be more exact to consider that leaves inhibit flowering under long daylength, rather than induce flowering under short days. An hormonal hypothesis for flower initiation is more likely than a trophic hypothesis[82] because etiolated plants still produce flowers. Auxin homologues stimulate vegetative growth and inhibit flowering, as ABA homologues enhance the effectiveness of short days to stimulate flower initiation.[80, 87] Accordingly, various hormonal schemes for flowering have been proposed.[82, 86]

The plant bears an increasing number of flowers for a few weeks.[40, 92] The definition of flowering date is rarely precise in literature, but is more or less related to the opening of the first flowers. Between flower initiation and flowering (i.e. some weeks), there is a progressive decrease in growth of stem DW but elongation of the very last internodes occurs with considerable reduction in the size of the last leaves. Flowering needs sufficiently high temperatures.[73, 91] A colder temperature can result in a return to vegetative growth[87] or evolution limited to buds only. Flowering also needs adequate radiation as shaded plants (the weak or smallest plants in a canopy) give fewer flowers, and too short days reduce or suppress flowering and relatively strengthen tuberisation.[48, 82] Suppression of *H. tuberosus* leaves grafted on *H. annuus* speeds up flowering.[89] Thus leaves have some role inhibiting flowering as well as against flower initiation. Plants kept for a long time in non flower-inducing conditions produce tubers, which will in turn hinder flowering. As a consequence, the harvest index is increased.[82]

Thus, there is probably competition between tuber growth and flower growth: prolonged short day treatment favours tubers against flowers.[80]

Flowers are often sterile and viability of the achenes is low and very cv-dependent.[67,91] Wild populations flower more and have a higher achene viability (up to 40%) than cultivated cv.[93] Consequently seed growth is generally weak even if earlier flowering results in more seeds being produced.[89] Irregular meiosis[89] and a strong auto-incompatibility system[88] explain low fertility of flowers. High temperature and a minimum temperature sum are required for fructification.[89] After maturity, seeds show strong dormancy or inhibition which can be suppressed with various treatments.[88]

4.3. *The relationship between flowering and tuberisation*

Any factor (osmotic pressure, antigibberellic, etc.) with an inhibiting effect on growth in length, induces swelling of stolons and tuberisation. These various factors have cumulative effects. Tuberisation would always be possible but is just expressed when growth is inhibited.[44] Changes in daylength and growth regulator balance are correlated with initiation of first tubers as well as induction of flowers. Nevertheless in late cv, if flower induction is followed by an increase in tuber number and growth rate,[37,78] first tubers are set before this induction. Flower initiation means a change in the hormonal status of the plant, as well as increased availability of assimilates because aerial growth is decreasing.[80] As a consequence, tuberisation could be induced by only the excess of assimilates relatively to the amount required for whole plant structural growth and existing storage processes, resulting in an excessively high sucrose level in stolons compared with their potential initiation and growth. This scheme is consistent with the appearance of first tubers before flower initiation and enhanced tuberisation after flower initiation in late flowering cv with relatively slow aerial growth.[78] It is also consistent with tuberisation just before flowering (and practically no slow phase in tuber growth) in early flowering cv with a high potential aerial growth rate. The balance between ABA and gibberellin (both deriving from mevalonic acid[74]), possibly altered by daylength (or flowering processes), would nevertheless explain differences in tuber shape through the inhibition of extension. This last process still reinforces sucrose concentration in stolons and assimilate-driven tuberisation.

5. THE CHARACTERISTICS OF FRUCTAN METABOLISM

5.1. *Characteristics of glucidic metabolism in* H. tuberosus

In *H. tuberosus*, starch is absent or rare, in leaves,[38,94] stems[27] or tubers. On the contrary fructans can be found in all organs of the plant, apart from all young growing tips. Metabolic pathways for carbohydrates, other than fructans or starch, are marginal.[38]

Fructans in *H. tuberosus* consist of linear fructosyl (F) polymers with a glucosyl (G) end.[38,95] The number of hexoses involved in the polymer is called the degree of polymerisation (DP). Theoretically, 'fructan' applies to a DP equal to or higher than 10, 'inulin' to a DP over 35. In mature tubers of *H. tuberosus*, most carbohydrates have a DP lower than or equal to 10, with an overall average around 5. Sucrose is the only carbohydrate form which is transferred within the plant,[96] and longer fructans are found in cell vacuoles only.[97]

Carbohydrate metabolism based on fructans has some advantages. It is less sensitive to low temperatures than other pathways.[38,98] The process of polymerisation–depolymerisation allows quick adaptation[38] of osmolarity and assimilate storage during growth (avoiding starch accumulation in leaves) and survival of tubers during winter (protecting cells against frost).

5.2. *Enzymology*

The first step in polymerisation is isokestose (G-F-F) synthesis in vacuoles from cytosolic sucrose (G-F). This involves the enzyme sucrose-sucrose-fructosyl transferase (SST) and co-produces glucose in the cytosol.[95] SST is probably linked to the system transporting sucrose through the tonoplast.[99] Glucose is then recycled to sucrose after phosphorylation involving sucrose synthase (SS) in the cytosol.[38,97] The equilibrium of this reaction is strongly directed toward G-F-F production.[95,97] A threshold level of sucrose is requested for SST activity, which applies to (and is regulated by) the cytoplasmic pool of sucrose.[97] SST is the key-enzyme for storage organs.[97] Its activity adapts exponentially within a few hours or days to substrate availability[97] and it disappears within a few days when tuber growth ceases or when the tuber is separated from the plant.

However it can be re-induced by sucrose.[95] Its activity is moderately sensitive to temperature.[97]

Further increases in DP involves fructan-fructan-fructosyl transferase (FFT), a vacuolar enzyme. It transfers a fructosyl end from a polymer to the end of another polymer or sucrose. Characteristics of FFT from *H. tuberosus* tubers have been described.[98] It is very insensitive to temperature and unable to hydrolyse G-F. Its transfer rate increases with substrate concentration. Affinity with F varies between polymers of different DP, maximum for sucrose and DP larger than 20, resulting in slow accumulation of polymers of higher DP.[95, 98, 100] A strong increase in triose is necessary before longer fructans are formed, because GFF is the best F donor (followed by short fructans).[97, 101] FFT is probably the only enzyme involved in polymer chain extension.[38, 96, 98] Distribution of fructans formed in the tuber is such that the amount of F is the same in each member of the fructan series.[101] During tuber growth, SST and FFT are the only active enzymes.[97] Contrary to SST, FFT is always present in tubers, but its activity undergoes some regulation as it decreases during the storage of tubers.[95]

Depolymerisation involves two kinds of enzymes. One is FFT because this enzyme modifies the distribution of DP when no more sucrose is provided to the organ. The other kind of enzymes are the fructan-exo-hydrolases (FEH), active on terminal F.[38, 102] FEHs are inactive on sucrose and non competitively inhibited by sucrose.[36, 97] Thus their activity is monitored by the release of sucrose. In resting tubers, only FFT and FEH are active.[97] FEH produces only F (for instance during sprouting[95]) out of the vacuole and it is linked to the tonoplast.[38] F released in cytosol is changed to G and then to sucrose by SS. The hydrolysis rate of FEH depends on the DP of substrate and increases with the DP up to 8, but is not related directly with fructose content or molar concentration.[102] Depolymerisation changes the DP distribution so that F is distributed equally (on a weight basis) among the members of the fructan series.[102, 103] Thus, during storage of tubers, a decrease of DP occurs without carbon consumption,[38] even at low temperatures.

To sum up, storage of fructans is positively regulated by sucrose availability through SST activity, and conversely removal from storage is negatively regulated through FEH activity. Fructans (triose and higher DP) accumulate only in vacuoles, and the anabolic and catabolic enzymes are on the tonoplast (with specific transport systems for mono and disaccharides) or within the vacuole.[97, 99] Mono and disaccharides are rare in the vacuole, most of them being in the cytoplasm.[38, 99] The cost of polymerisation is estimated at 4%–8% of the mass of assimilates,[60, 78] most of this cost being related to the recycling of glucose to fructose in the cytoplasm. Almost all enzymological studies have been done with tubers. Their extrapolation to the storage mechanism in stems is hypothetical, but it is noteworthy that the same series of compounds are found in all organs.[15]

The accumulation of polymers (DP > 5) is faster and greater in tubers than in stems and faster for high polymers in young than in old tubers.[96] Assimilates are stored as short polymers within a few hours and within a few days for higher polymers (1–3 days for DP > 7).[96, 104]

5.3. *Consequences for quality and uses of sugars*

During the period of strong growth of tubers, the percentage of long DP fructans tends to increase, even if the average DP can temporarily decrease because of the strong influx of sucrose resulting in large amounts of G-F-F.[27, 36] The highest average DP is reached some time before the maximum DW of tubers. At the end of transfers of reserves from the stem, the average DP is around 10. Then, in mature tubers, activity of FFT and FEH results in rapid changes in distribution of DP toward a lower average value (from 8–10 to 4–6, with an increase in G and a decrease in F).[17, 40] Consequently tubers become more fermentable as they age, thus more suitable for alcohol production and less for fructose production because of the relative loss in F.[53, 105] The degradation of high DP (> 10) polymers is nearly complete within a few weeks whatever the temperature. DW and DP are often found to be higher in late cv than in early cv.[40, 56] This is, at least, partly the result of a common harvest date with more aged tubers for early cv. For similar physiological ages, the F/G ratio is very similar between cv.[39]

Losses of sugar during storage have been quantified at 1% to 2% per week[45] and sometimes less (0.5%–0.7% /week)[17, 30] when tubers are kept in the soil. In this last case, losses can be underestimated because of the disappearance of rotten tubers. Moreover, depolymerisation means uptake of water (as H plus OH) by molecules during hydrolysis. As a consequence, the DW can increase during winter. Thus

respiration, and losses, can be underestimated if only DW is considered without correction for F and G equivalents, involving a change of DP.

6. DISTRIBUTION OF GROWTH AND REALLOCATION OF COMPOUNDS

6.1. *The growth of aerial structures and roots*

Stems and branches represent the largest part of the aerial DW (2/3 to 3/4). The rest is mostly leaves. Flowers, depending on cv, represent only a few percent.[50] Stem number per plant depends on variety,[27,75] seed-tuber size,[27,75] and tuber-ageing.[75]

After the juvenile phase with opposed phyllotaxy,[57,82] leaves then become set in verticils of 3 or on a helix and internodes become shorter.[45,52] Internode number is more constant in early than in late cv,[45] and is reduced with increasing planting density.[77]

Distribution of DW along the stem shows a predominance with lower internodes, and large variations with time.[27] Elongation of the stem responds strongly to water availability.[8] Flower initiation stops the growth of aerial parts, resulting for instance in small plants and short duration of growth under tropical conditions probably because of short daylength. All aerial structural matter is formed a short time before flowering, when the aerial DW is at its maximum.[12]

The number and size of branches are varietal characters, strongly influenced by growth conditions, with a strong inverse correlation with planting density.[49,77] Branches grow later than the main stem, and the higher branches much later.[106] Until flowering, they grow mostly at the base of the stem.[52] The onset of flowering induces numerous small terminal branches bearing flowers, because of the loss of dominance of the terminal bud.[40,48]

Competition for growth between aerial organs has been reported in numerous conditions, explaining the high morphological plasticity of this crop. Suppression of one branch enhances growth of the closest leaf and the weight of the stem.[106] Suppression of one leaf (or one leaf with its associated branch) reduces growth of the subsequent internode but induces stronger growth of (mostly lower) branches.[106]

The growth of the subterranean stem is parallel to the growth of the aerial stem.[75] An early treatment with MCPA results in a transient increase followed by an early cessation of stem growth.[87] Treatment with triapenthenol reduces stem length somewhat.[3,78]

Many authors report the considerable ability of the crop to colonise soil and extract minerals but studies on root system growth are very rare.[45] *H. tuberosus* is not tap-rooted like sunflower, but fasciculated (bunchy)[52] with a rooting depth of more than 1 m.[3]

A high planting rate results in, at least temporarily, taller stems and increased elongation rate. At a very high density the final size of stems is still shorter.[57,77] High planting densities also decrease stem diameter more than stem height.[77] Increasing plant density increases the total DM production, DM percentage of tubers and harvest index but decreases the number of tubers per plant.[56] The number and quality[53] of first order tubers are not changed,[77] but they are smaller.[8,77] High to very high planting densities reduce the growth in weight of stems, and more the growth of leaves.[57,77]

6.2. *Storage of temporary reserves*

Nearly all organs of the plant can store some reserves, at least temporarily. Variations in the reserve content (carbohydrates and proteins) with time and relative sampling date explains most of the contradictory conclusions between references referring to the quality of aerial parts.

Upper leaves store assimilates (mostly fructose and sucrose, few fructans, some starch)[27,38,94] on a daily basis; thus their reserve content varies greatly during a day[27] and is low[17] in the morning.

There is often some confusion in the literature between true roots and the subterranean stem (sometimes called 'stump'). Roots are also able to store and polymerise fructans, even later than the stem,[96] and their DW decreases during final tuber growth. Up until this time, they can contain more than 40% of non structural carbohydrates[30] and they are able to store large amounts of sugar if the tubers are removed.[60] They can thus be considered as a storage organ with low priority level.

The percentage of sugar reserve in stems varies greatly between references, depending on the time of year, year, location and varieties.[16] At its maximum, the sugar reserve can be as high as 25% to 70%[30,39,88,107] of DW, or 14%–17% of FW,[17] and somewhat higher (but more variable) for early than for late cv.[39,107] During vegetative growth only one-third (maximum) of assimilates is transferred to growing apices.[96] Thus there are assimilates available for growth

of lower parts and storage. This last process relies on assimilates in excess for structural growth, as shown when suppression of a large part of the leaves reduces the stem DW per unit fresh volume (and thus probably stem reserves), and tuber growth.[108] Reserves are stored in the stem, in parallel with tuber growth, at least until flower initiation,[78] flower bud formation,[3] or even until[17, 107] or some time after flowering.[54]

During the storage of reserves, the percentage of sugar and average DP increases more towards the bottom in the stem.[27, 94] Even when tubers already exist and slowly grow, sugars accumulate in the stem,[55, 96] mostly in the medullar part.[94] Transfer from phloem to medulla is stopped or very slow when the growth of tubers is strong.[60] As a rule, transfers are faster downward than radially in the stem.[96]

At flowering time[55] or a little earlier[96] there is hardly any transfer of assimilates to the upper stem apex. Remobilisation of reserves begins at the lowest part of the aerial stem, as expressed by the reduction in DP throughout the stem but with lower DP towards the bottom, except for the stump.[94, 107] Around 30% of DW is lost by the stem until the moment when tubers are at their maximum weight.[50] In early cv most reserves are transferred before foliage senescence; in late cv remobilisation still goes on after death of the foliage.[50] At the end of the transfer, no more (or very little) sugar is left in the stems.[26, 107] A low correlation between aerial DW and tuber yield[49, 64] is an expression of variability in assimilates and reserve distribution, with a possible reduction in the harvest index if the aerial structural growth is too strong.[51] Accordingly, accumulation of fructans is enhanced by high radiation, low temperatures, but lowered by an excess nitrogen or potassium deficiency. Strong aerial growth increases the hardiness of the plant but temporary storage of reserves in the stem nevertheless represents some waste of energy and carbon. At least 3 to 6 ATP/mole sucrose (4%–8% of its mass) are still needed for storage-destorage and additional transport, and storage tissues represent an additional material and energy (respiration) expense.[78]

6.3. *The growth of stolons and tubers*

Stolons never represent a large part of the plant dry weight, but they polymerise fructans very efficiently and accumulate a high percentage of sugar even when they are still growing.[96] At high planting density their appearance is postponed and their number per plant, their length, node number and amount of branching are reduced.[77] Thus stolonisation is probably driven by assimilate availability per plant. Branching of stolons proceeds until the time of flower bud appearance. At this time, mostly secondary tubers are formed and no more stolons. A third wave of tuberisation can occur at plant senescence[76] (remobilisation of stem reserves).

A high planting density does not change the tuberisation date.[77] With very high density, no stolons or tubers are formed, but roots or the lowest part of the stem can survive and accumulate reserves when the foliage dies.[77] Tuber number varies with time, because of progressive initiation. It is more or less correlated with a cumulated temperature since emergence.[65, 78] The final number of tubers, a varietal character, is sometimes found to be fixed at flowering, or still changing subsequently.[34]

The number of tubers per stem decreases with an increasing stem number per area, planting depth, or with late planting.[14, 75] Tuber weight per plant or stem is reduced by higher planting density or numerous stems, but weight and number per area are less, and often inversely, modified.[8, 77] As a consequence, increased planting density often results in increased tuber DW. Within a cv, total tuber yield is loosely correlated with tuber or stolon number.[65]

Distribution of the size of tubers is an important parameter for harvest: losses at harvest can be as high as 15%–20% DW; tubers smaller than 15 g FW cannot be harvested.[4, 43] Tuber size distribution is still rarely described[49, 109] or explained. The number and average size of tubers are generally negatively correlated. Monostem plants are thus preferable because the increase in stem number results at its best in slightly higher potential yield, but with more and smaller tubers[8] inducing more losses at harvest.

The number of tubers depends on assimilate availability.[6, 65] Growth factors may also be involved, as various types of suppression of organs results in different modifications in the number and size of tubers,[106] and short day treatment increases tuber number.[80] The apparent effect of photoperiod on tuberisation is also related to flower induction and the consequent decrease of aerial growth.[60] Tubers grow by utilizing current photosynthesis and by remobilisation of reserves from various other plant parts.[8, 36] The first source takes priority over the second.[26, 57] The correlation between dry weight

of stems at specific dates and tuber yield is generally weak.[49,64] Thus the reserve percentage in stems is not fixed and, accordingly, part of tuber growth on reserve remobilisation is variable. Low remobilisation (for instance, in case of frost) is one of the reasons for low yield with late cv in high latitudes.[109] Transfers from stems are stopped when air temperature drops below $-3°C$[51] but can recover for some time afterwards.[45] In early cv the tubers grow with a nearly constant rate,[49,109] but in late cv they grow slowly until the beginning of flower initiation and then far stronger after some lag time.[78,88] Carbohydrates, amino acids and minerals are transferred to tubers,[53] sucrose being the only type of carbohydrates[60]; the transfer from stems to tubers (estimated from stem DW loss) is up to 50% of final tuber DW.[39,107] Nevertheless an unexplained large part of the fructans lost by the stem is not gained by tubers.[78]

During the period of maximum growth, the growth rate per tuber (DW/time) is almost constant[30,78] and is thus supposed to be sink limited.[37,78] The increase of tuber number with thermal time would explain an apparent correlation of the growth of the whole tuber set with temperature.[37,78] The final weight of a tuber is nevertheless correlated with the earliness of its formation.[76] During tuber formation, the xylem expands considerably, but actual growth of tubers is limited to parenchyma (particularly cortical parenchyma).[60,72] Cell division, growth and storage activity are simultaneous in a tuber.[60] Fresh sugars accumulate in young growing peripheral cells and there is little or no exchange with older internal cells.[60] Growth in diameter does not stop growth in length.[75]

6.4. *Competition for growth at plant level*

There are various expressions of competition for growth between stem and tubers. Aerial parts and tubers can both be used but their uses are mutually exclusive. Harvest of the green aerial parts strongly reduces tuber yield[8,14] by preventing remobilisation. A related question is the mechanism of assimilate and reserve distribution within the plant.

Suppression of buds, apices, flowers or flower buds, by suppressing concurrent sinks, promotes tuberisation (greater increase in individual mass than in number) without change of mass of stem and roots nor change in total biomass.[44,93] The difference in tuber yield between various cvs of similar lateness mainly depends on the percentage of growth devoted to flowers and stem with total biomass being very similar.[93] Accordingly, defoliation reduces tuberisation and increases internode length in stolons in lowering assimilate supply.[44] In the case of severe temporary defoliation, stem and foliage regrowth have apparent priority over tuber growth, but stolons and tubers (even strongly reduced) are nevertheless produced.[67] Treatment with growth inhibitors like triapenthenol or other triazoles (inhibitors of synthesis of gibberelic acid) results in a decrease of stem length, increase in tuber number and mass, and sometimes increased tuber sugar content.[3,78] On the contrary, late treatment with an auxin homologue extends vegetative growth and results in increased water consumption and decrease in sugar content of tubers.

Thus, treatments at crop level confirm the conclusions drawn *in vitro* where any inhibition of growth or elongation results in tuberisation (see above) and vice versa. In the field, sometime after flower initiation, a reduction in growth potential of the aerial parts results in enhanced growth of tubers.[78] Tuber growth is also promoted when the assimilate level is increased in the plant, because of stronger radiation or mild stress reducing structural growth more than photosynthesis.[88] As a rule, tuber growth can be seen as a response to assimilate availability in the case of reduction in aerial growth.[60] If tubers are removed when they should normally grow, sugars accumulate in the stump and roots. Thus there are always other possible sinks for storage, with a lower priority level, while the final storage is source-driven.[60] Similarly, when growth conditions are very good, the lateral buds of tubers are no longer dominated, resulting in tubers with very irregular form.[52] There is also competition between tubers, as the biggest tubers get a larger fraction (more than proportional to FW) of assimilates.[60] Aerial growth (buds) has, at least temporarily, priority over tubers, and tuberisation can be seen as an overflow for sugars when the stem cannot use or store them any more.

In contrast, labelled ^{14}C provided to plants with tubers is found in growing parts of tubers, and only as sucrose in the vascular tissue of the stem; it is not found in stem pith, and transfer to roots is very low.[96,104] Storage in existing tubers thus has priority over storage in stem. Very high planting density reduces tuber growth more than aerial growth, and can even suppress tuberisation.[57] Thus, a 'minimal' aerial growth has priority over any storage process. When

photoperiodic-response plants are kept at their vegetative stage with artificial light, they nevertheless transfer reserves from the stem to tubers in autumn but later than control plants.[78] In these conditions, cold air temperatures could lessen aerial sink strength. Partial defoliation reduces stem and tuber growth, and decreases the dry weight of stem per unit volume.[108] When similar defoliation is combined with suppression of the stem apex, tuber growth and stem diameter are increased close to control.[108] Structural growth of the stem thus has priority over reserve storage in stems and tubers.[108] Shading experiments result in fewer reserves in aerial parts but no reduction of tuber growth.[66] This shows that storage in the stem has less priority than tuber growth and can be seen as an overflow for assimilates when tuber growth is limited by their own sink capacity. To sum up, the priority order at the whole plant level seems to be: aerial structural growth, then tubers (when existing) and finally stem reserves. Thus tuber growth is limited on the one hand by the source capacity of total growth minus structural growth and on the other hand by its own potential growth. Tuber initiation depends on assimilate availability at the whole plant level (cf. Section 4.3). At least one question nevertheless remains unanswered: why does the stem storage system become less competitive after some time than the tuber system?

In existing models, distribution of growth between plant parts has been described empirically as a function of time or of temperature sum,[28, 57] or as resulting from competing sinks with hierarchical relations[28] but with empirically fixed size. Until now, the mechanism of assimilate distribution among aerial structures, aerial reserves and tubers has remained hypothetical. Consequently, simulation remains less satisfactory for tuber yield than for total plant growth.[29]

7. THE ROLES OF GROWTH LIMITING FACTORS

7.1. *Weeds, diseases*

H. tuberosus is considered as a very hardy crop and already had this reputation when it was much more widespread than now and thus more exposed to pest epidemics.[8]

Except during crop establishment, the competition by weeds against *H. tuberosus* is negligible.[1] Some time after emergence, earthing-up and hoeing 2–3 times are sufficient to control weeds and build ridges with a light structure which is adequate for tuber growth. Chemicals can nevertheless be used to control weeds before crop emergence,[51, 53] or afterwards until canopy closure,[43, 50] but mechanical weeding (earthing-up, hoeing, etc) is preferable.[53, 55]

Diseases are common on tubers during late growth and above all during storage,[35, 53] but low temperature and fungicides can control the damage.[3, 110] Diseases are much less numerous and less dangerous on aerial parts and during crop growth.[92, 110] Mildew (*Erisyphe chicoracei*) is the most common disease but has a low impact.[53, 92] *Sclerotinia sclerotiorum* (promoted by excessive N fertilisation, low pH,[3] or hydromorphic soils[8]) is more harmful as it totally destroys the plant, but fortunately this disease spreads at a low rate in the field.[35, 51] Thus it is dangerous only after some time, and can be controlled with chemicals[3, 110] or appropriate crop rotation.[110] *Sclerotinia rolfsii* (found only in north America) causes heavy damage; it is favoured by abundant rain and high temperature.[110] Chlorosis is sometimes recorded in America and is ascribed to *Pseudomonas syringae* pv *tagetis*. It can be transmitted through seed-tubers, but it causes transitory and very limited damage in crops.[3] The rust *Puccinia helianthi* is also sometimes recorded; its impact is very variable and it can be controlled chemically.[92, 110]

Aphids are common, and the sunflower stem maggot (*Strauzia longipennis*) is found in America.[92] As with other insects,[53] their impact is practically negligible. Because of the long duration of the crop in the field, some losses (particularly in early spring) can be incurred by game (deer, rabbits, etc).[8, 12] *H. tuberosus* is a non-host plant for soil nematodes.[3]

7.2. *Water*

H. tuberosus is more hardy against drought than many other crops, but it is sensitive to excess water.[8]

Morphology is modified by drought: plant height (indeed, length of internodes) is reduced and can vary considerably.[8, 45] LAI is sensitive to water shortage[55, 111]: specific leaf area is increased resulting in slower LAI increase, and leaf life span is shortened, resulting in earlier foliage senescence.

Water stresses reduce aerial and subterranean growth,[112] and generally tuber yield.[55] Nevertheless the harvest index is often improved,[45] because of a relatively strong reduction of aerial

growth,[37] resulting sometimes in increased tuber yield.[105] Nevertheless irrigation is generally worthwhile when available.[49]

Tuber yield is highly sensitive to drought during late tuber growth[45,49] or around flowering.[111,113] As a consequence, early cvs are more affected than late cvs.[3,88] At this time photosynthesis and transfer of reserves to tubers (and stem senescence) are hampered and thus can explain the yield reduction.[45,72] Final tuber bulking can be limited by water supply.[17] Emergence is the second sensitive stage.[49] Stress during early vegetative growth is less damaging for final yield than during any other stage.[111] Continuous slight water limitation, or limitation before flowering somewhat reduces (or sometimes increases) tuber yield but improves water use efficiency (WUE) and HI, contrary to late or very strong stress.[111,113] Water use efficiency is poor anyway, because of poor stomatal regulation.[30,57,111] Tuber number is affected by drought but with unexplained and inconsistent patterns.[43,112] Crop coefficients for maximum evapotranspiration have been calculated.[111]

7.3. *Nutrients*

The effect of mineral nutrition is generally analysed poorly. Information about initial soil conditions or mineral balance is generally lacking.[114] As a consequence, many contradictions occur in the literature and recommendations for fertilisation vary strongly between references. The uptake rate is highest during summer months and maximum uptake is at the time of maximum total DW of the plant.[43,50] *H. tuberosus* has a strong ability to extract nutrients from the soil,[8,114] particularly potash and phosphorus. Good yields can therefore be obtained for years with very low fertilisation[53] at the expense of existing soil reserves, as nutrient exports are often higher than supply.[12,14] As a consequence, yield responses to P and K are often low.[114] However the crop still needs large amounts of potassium,[35,43] nitrogen[43,50] and calcium[43,50] for an optimum yield, even if the final mineral export[14,53] is lower than the maximum content of a standing crop. Tubers export 51%–57% N taken up, 58%–81% for P, 36%–48% for K, 6%–8% for Ca and 14%–24% for Mg.[50]

Lack of P or K disturbs tuber morphogenesis, growth and yield, more than aerial growth.[114] The yield response to nitrogen is nevertheless stronger than to potassium, probably because of the difference in their original content in the soil and because nitrogen determines potential photosynthesis and somewhat increases water use efficiency.[64] High nitrogen fertilisation can result in over-consumption, excessive aerial growth and consequent decrease in harvest index and tuber yield.[8,43] Thus mild nutrient stress could improve yield.[34] Information on the effect of macronutrients on micronutrient uptake is scarce.[49,114] Some necrosis, without pathogens, of the upper leaves during warm periods[51] could be because of trophic imbalance.

Yield response to fertilisation seems to depend somewhat on cv.[114] This could be the result of differences in potential growth inducing different relative shortages, as there are few differences between cv of similar lateness with regard to mineral immobilisation or export.[3] Depending on cv, fertilisation affects mainly the number or size of tubers[114] but has no effect on sugar quality[55,114] (DP of fructans...).

The P, K, Mg, Cu and Zn content of tubers depends slightly on fertilisation and often decreases when fertilisation is increased, probably because of greater dilution. Ca, S, Mn and Fe levels are unchanged; on the contrary, the N content is slightly positively correlated with fertilisation.[50,114]

7.4. *Other factors*

During its early growth, the crop is not very sensitive to frost,[3,8] even if cold temperatures can induce foliar chlorosis.[73] Young shoots can withstand temperatures as low as $-6°C$ (soil level) without damage.[52] A standing crop of a late cv of *H. tuberosus* can thus overwinter in mild climates and give a higher yield.[35] The crop is more sensitive to autumn frost[3,52] but some cv can withstand (remain green) -3 to $-3.9°C$ night temperatures. Others are more sensitive,[30,52] probably depending on soluble reserve levels in the leaves.

Tall stems can be broken or the foliage can be destroyed by storms.[51,109]

The crop is moderately tolerant to salinity, with a poor control of Cl accumulation, and tuber yield is moderately sensitive to salinity when the soil is heavy.[71] Emergence, plant height, branching and LAI development are the most sensitive to salinity, unlike tuber mineral content.[71]

8. DISCUSSION

From the information already available on *H. tuberosus*, it is possible to establish some

directions for building dynamic simulation models and to identify gaps in knowledge for this task.

8.1. *Vegetation establishment and duration*

Any factor favouring early growth and a quick closure of canopy increases potential growth through an increase of photosynthesis in Spring at the crop level. Dormancy is practically always broken in Spring, and would only need breaking in the case of early planting (Autumn, early Spring) in temperate climates. Modelling the duration of crop life needs a model of crop emergence (and plant losses at emergence) depending first on temperature and secondly on the moisture regime of the soil. Precision is needed for the date of emergence because early growth is nearly exponential. Death of foliage is the limit for total biomass accumulation; it depends on a model for LAI growth and senescence. Some investigation is needed about the rate of transfer of reserve from stem to tubers, and its possible limiting factors (cold temperature, water stress). This transfer of reserves and consequently tuber growth can indeed go on after foliage death but could be hampered by adverse environmental conditions.

8.2. *Dynamics of leaf area index (LAI) and light interception*

As duration and efficiency of canopy photosynthesis is crucial to determine the total productivity, a robust model of the dynamics of leaf area index, or light interception, is needed urgently. It will also have to rely on temperature for potential foliage growth until flowering. Seed rate and seed-tuber size while having some effect on stem number are second order parameters, improving the simulation until canopy closure. Foliage senescence should take into account the assimilation–respiration balance of the canopy layers and the dynamics of concurrent sinks, particularly for nitrogen. Research is needed here on respiration, N gradients within the canopy (to estimate potential assimilation), and the N requirements of competing sinks. Leaf area index measurements are rare, and the estimation of dead LAI is non-existent. Data are nevertheless necessary for parametrisation of a complete LAI model. Simpler but less mechanistic models of evolution of light interception can alternatively, and at a lower experimental cost, be improved for wider relevance, for instance with better insight to responses to temperature.

Various light interception models have been tested already, and their parameters estimated for a few cvs.[25, 28, 37, 62] As the canopy requires some weeks to close, introduction of a row effect on light interception should be investigated to improve the simulation of early growth.

8.3. *Photosynthesis and total growth*

Information about the photosynthesis of *H. tuberosus* is scarce. Total productivity of a standing crop (at least, when expressed as assimilates) can be nevertheless modelled the same way as for various C3 crops without details about photosynthetic processes. Nevertheless, dependence of photosynthetic assimilation on extreme temperatures, high photon flux density, and leaf age needs to be investigated to better assess productivity of the crop in relatively warm regions where it does best. Maintenance respiration of various organs is poorly known and would represent a weak point of any model including carbon balance. Measurement of this process and its dependence on temperature is needed for comprehensive models. Alternatively, a direct expression of intercepted radiation as equivalent biomass assumes a chemical composition and is less precise than an approach based on actual chemical composition and including the effect of temperature on respiration. A direct equivalence is still simpler to establish and reasonably accurate insofar as chemical composition does not vary too much.

8.4. *Chemical composition*

Stage and age of organs (and perhaps variety) are important in determining their chemical composition and are not always fully explained in the literature. As a consequence the values in the existing literature must be used with caution. Definition of the total N content (and calculated protein content insofar as no inorganic nitrogen is stored) is generally clear. In contrast, the estimation of fructans is far more dubious: procedures for extraction, depolymerisation, and sugar determination are not standardised and are never compared with one another. For instance, fructose and glucose are generally quantified after hydrolysis but correction for weight gain at hydrolysis is missing (or not cited). This simple omission induces an error of 5%–10% on fructan DW, depending on the degree of polymerisation of the sample. The very high ash content of some organs (leaves) also requires some corrections to be expressed

as minerals. Methodological investigations are thus needed to reliably compare existing data and to define future methods. Additionally, standard coefficients relative to transformation of assimilates to biological compounds and tissues can be used, because most chemical compounds in *H. tuberosus* are common. Biochemical work is nevertheless needed to better estimate the energetic cost of storage and transfers of fructans.

8.5. *Fructan metabolism*

Fructan metabolism allows quick storage of large amounts of assimilates in vacuoles of living cells. This metabolism has never been studied in organs other than tubers, it is still hypothesised that the enzymatic mechanism is the same in all plant parts. This point needs experimental confirmation.

Fructan metabolism is regulated by sucrose availability. This allows the plant to adapt its growth to the potential determined by photosynthesis, within the limits of the maximum activity of existing (induced) enzymes. The sucrose influx to the cell also regulates the degree of polymerisation (i.e. quality) of fructans. The enzymatic system is directed towards storage of sucrose as vacuolar fructans, as long as cytosolic sucrose is above a certain threshold. It is inversely directed towards the gradual release of sucrose (because of negative feedback on FEH activity) in the opposite case.

A typical and unexplained trait of *H. tuberosus* is that its stem is first a storing and then a releasing organ, in spite of its position close to source organs (leaves). A change in the balance of growth regulators has been hypothesised to explain this, but it is unclear how the effect of growth regulators would be different between two organs (i.e. tubers and stem) with the same enzymatic system. Some additive role of light can be supposed, since darkness promotes storage and tuberisation, but it has never been studied. Another hypothesis is worth some investigation to explain fructan management in a whole plant: following the basic principles of enzymatic activities and their equilibrium at the cell level, one may suppose a decrease in sucrose pro- cessing capacity or 'sink strength' of a storing cell when its reserve content increases. As a consequence, the ratio reserve sugar/DW tends asymptotically towards a maximum value and simultaneously the sink strength tends to zero. Thus, changes in storage activity of the stem can be explained as a type of saturation of storage potential. Later release of reserves by the stem is a consequence of the appearance of the tubers as strong competing sinks with active cell division (contrary to a flowering stem) inducing an increasing sink capacity, whereas the pro- gressive export of sucrose from the stem does not allow the re-induction of SST activity in this organ.

8.6. *Development*

Flower induction is mostly driven by daylength in late cv: a given number of days under a threshold daylength value determines flower initiation in field conditions. In early cv, investigations are still necessary to explain induction since temperature as well as plant growth seems to have some role. Whatever the earlyness, flowering and fruiting depend mostly on temperature and also growth. Flowering and fruiting are however of low impact on crop yield and quality. In addition to research on flower initiation in early cv, experiments are also needed to estimate the cv-dependent parameters of flowering (from initiation to fruit). Precision is needed because flowering is a major shift in crop physiology.

Stolonisation and tuberisation can be considered as mostly growth-driven phenomena. Assimilates are required and sufficient to induce tuber initiation and growth whereas, in the present state of knowledge, this is not the case of any growth regulator. Growth regulators have a probable additive role in restraining stolon elongation thus favouring the swelling of stolon tips as tubers. Both causes are related to flowering because flower initiation changes the hormonal balance of the plant and reduces the aerial parts' requirements for assimilates.

Indeed, no experiment has indisputably proven any hypothesis, but the following scheme is the most consistent with the whole experimental *corpus*: stolonisation occurs when assimilate supply is sufficient for resting buds on the subterranean stem, and subsequently, on stolons. When the potential growth of existing organs is fulfilled, new stolons can expand and accumulate sugars. Over a threshold level of accumulation (perhaps in stolon tips, when sucrose influx takes over potential catabolism), stolons swell to become tubers through multiplication of cortical cells and reduction in the length of the internodes. Growth in stolon length is still possible and consequently their shape is not typical. As a result of competition with other plant parts (growth of aerial

structure, storage in stem), tuber growth is slow as long as competing sinks are not repressed (for instance, with chemical growth inhibitors or under cold temperature) or saturated. When flowers are initiated, the size of vegetative sinks competing with tubers is fixed. Since the branches initiate flowers somewhat later than main stems, the number of branches would need to be introduced in calculating final structural growth. The amount of reserve tissues in stems depends on structural tissues and is consequently limited. The aerial storage capacity is progressively fulfilled and assimilates become increasingly available for tubers. At the same time (perhaps, progressively) the growth regulator balance changes (ratio gibberellins/ABA?) in relation to flower initiation. Repression of elongation results in stimulation of the cortical growth of tubers. The active storing tissues of tubers increase their mass and become as a whole more effective than the stem. As tubers are also better connected to phloem vessels, they are better able to unload assimilates from the phloem and then reserves from stem cells. Standby of storage activity in stem results in the disappearance of SST in stem cells and the consequent disappearance of a corresponding sink strength for sucrose.

Of course, this scheme still needs confirmation. *In vitro* experiments (organ culture) or artificial phloem loading would provide evidence of induction of stolonisation or tuberisation with assimilates. As a first stage, a simulation model could be used to test the whole scheme and compare its behaviour with detailed growth analysis data.

For further simulation purposes, growth regulators can be explicitly disregarded because they act mostly as transmitters of environmental stimuli. Moreover, the present state of knowledge does not make it possible to simulate their amount or concentration and these data are difficult to obtain experimentally. On the contrary, the amounts of biomass and assimilates are easily measured and simulated. Within the scheme of integrated plant growth proposed here, stolonisation and tuberisation depend on potential rates of structural growth and storage in competing organs. In the literature, data are nevertheless lacking on these points.

8.7. *The structural growth of the plant*

Temperature is probably the most limiting parameter for structural growth. Definition of the 'effective temperature' is therefore urgently needed as a main driving variable. Assimilate availability is probably never limiting, as the seed-tuber supports early growth and the plant can already store reserves very early in its life. Potential aerial growth seems to be unlimited until flowering because of the potentially strong branching which is still regulated by plant density. There are nevertheless numerous growth correlations and internal competition at plant level. Relationships between organs have been rarely qualitatively (and even less quantitatively) explained or described. A model dealing explicitly with all relationships between the numerous organs would probably be too complex and unmanageable. Moreover, many growth functions are positively or negatively correlated to one another: branching compensates for insufficient stem growth, subterranean growth is related to aerial stem growth, etc. Consequently, complete aerial growth could be, in an initial approach, simulated with empirical functions of effective temperature. Investigation of growth correlations (allometric relations, etc.) would help to provide a better insight into morphogenesis of the whole plant and reduce the amount of parameters needed for a model. There are very few data to quantify structural growth because stem reserves have been rarely separated from the structure. Moreover, structural growth is sensitive to slight limitations of water or nitrogen availability. Consequently, experimental efforts are still needed to gather consistent data in well defined conditions. At the plant population level, the role of planting density must be explained to give some insight into experimental variations in harvest index.

8.8. *Reserve storage and the growth of storage organs*

As a rule, storage of reserves is the consequence of an excessive assimilation relative to the potential structural growth. Apparently (but still to be proven), photosynthesis never undergoes any regulation because of a lack of a potential sink. Storage first takes place in the stem and branches because they are closer to leaves and because storage tissue (parenchyma), created at the same time as structural tissues, is available from the beginning. Then storage begins (apparently, more effectively) in stolons, which are nothing other than subterranean branches. The scheme proposed in Section 8.6 applies to reserve management: development of subterranean parts is here a consequence of growth distribution (or overflow of assimilates).

Some structural growth (cell walls, etc.) is nevertheless needed for tuber growth. Investigation is necessary to know to what extent the rate of tuber initiation or structural growth can be limiting for reserve storage, as tuber growth often appears to be 'sink limited'. Such limitation could also be the result of the maximum processing capacity of tubers (enzyme saturation), or the maximum potential transfer rate (size of sieve tubes, water availability, sieve quality). Better insight into this question would make it possible to better quantify the tuber growth rate. Accordingly, the number of tubers and distribution of their size, which is important information for determining harvestable biomass, could also be simulated. Within a scheme of assimilate-driven tuberisation, it becomes possible to simulate 'cohorts' of newly created tubers (with a fixed individual size) according to daily overflow of assimilates, and then growth of each cohort.

8.9. *Growth limiting factors*

Water availability is likely to be the most striking growth limiting factor, as the crop withstands pests and diseases and efficiently extracts nutrients from the soil. Moreover, growth of aerial parts (leaves and stems) is the most sensitive process to any water shortage. It indirectly influences the storage of reserves and tuber growth if structural growth is more affected than photosynthesis, as is seen when mild water stress results in increased tuber yield. The transpiration regime of the crop and role of water status in structural growth therefore needs to be studied urgently for later modelling of growth under water limited conditions. The hardy *H. tuberosus* would indeed probably be planted in such conditions. But information on water balance at the crop level and on yield is very scarce. Collection of data is urgent for a first approach to this question. Nitrogen is the second limiting factor, with a role in aerial growth and perhaps growth distribution. The coupling of a crop model with a soil model (for water and nitrogen) would, however, need improved knowledge on plant rooting dynamics.

8.10. *Crop qualitative parameters*

Quality of plant organs depends mostly on fructans and protein content. Until now, they have been empirically correlated with approximate development stages but with poor precision. Quality can be simulated as far as non-structural carbohydrates and nitrogeneous compounds are quantified in each category of organs in a model.

In tubers, the N content seems to be rather stable, allowing some simplification of a model, but also inducing some constraints on carbon flows (C/N ratio). The amount of fructans depends on the scheme for reserve management presented earlier. DP of fructans could be related to the history of carbohydrate flow in the organ, but data are still very incomplete regarding this point. An integrated and quantitative model of the fructan-enzymatic system at cell and organ level is needed to simulate evolution of various pools of carbohydrates. In spite of recent efforts,[96, 97, 100] there is still much to be done here to integrate knowledge and validate a model.

Mechanisms inducing variation of content in N and minerals of aerial parts are still unclear to a large extent. General schemes for N dilution, defined for C3 plants, could be tested against available data. The mechanism of response of aerial growth to excessive N fertilisation needs further investigation for a more mechanistic determination of optimum fertilisation. Similarly to the case in tubers, the amount and characteristics of fructans in stems could be simulated within the scheme proposed in Section 8.6 for tuberization and Section 8.8 for assimilate management at the plant level if quantitative relationships are established between fructan metabolism and sucrose influx at organ level (Section 8.5).

9. CONCLUSION

In spite of limited research devoted until now to *H. tuberosus*, much factual information is available, often in old literature. There is nevertheless considerable methodological concern about using directly data from the literature, because of the variability in measurement and analytical methods.

Modelling may be helpful to structure this information and make it usable for estimating potential yield and quality. Directions for future research, which is adequate for modelling, can be also derived from analysing the literature. The order of priority amongst these model parameters is a matter for conjecture. However, for modelling purposes, basic questions on productivity (LAI dynamics, total growth) and development (emergence, flower initiation) must be answered first as subsequent steps depend on them.

Models may also be helpful to test the realism of some hypotheses, such as those dealing with assimilate distribution. Gaps between simulation and experiment may help to identify weak points in knowledge and better define future experimental work. In contrast, a total absence of fit or divergence from the model may invalidate a scheme.

Results obtained up to now in modelling yield of *H. tuberosus*[25, 26, 28, 29, 34, 62, 65] are encouraging and warrant more effort for a better understanding and applied simulation of the physiological behaviour of this crop.

Acknowledgements—The author is grateful to Pr R. Rabbinge, Dr R. Bonhomme and Ir W. Meijer for their valuable comments on the manuscript, Ir M. Pavlides for her thorough help in gathering documentation, Dr N. Ivanov for his help in exploiting references in Russian and Dr A. Fuchs (Agricultural University Wageningen, Dept Phytopathology, Wageningen, NL) in sharing a part of his unique reference base on inulin and inulin containing plants.

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